

**PROTOCOL TITLE:**

***Transcranial Magnetic Stimulation for Depressed Adults***  
***with Autism Spectrum Disorder***

**PRINCIPAL INVESTIGATOR:**

**McLeod Frampton Gwynette, MD**

*Do not include local co-investigators—they will be listed in the eIRB application under study personnel*

## 1.0 Objectives / Specific Aims

**Autism Spectrum Disorder (ASD) is a common neurodevelopmental disorder in the United States and globally, characterized by impaired social communication and interaction, along with restricted interests, behaviors and activities[1]. Additionally, individuals with ASD are at increased risk for multiple psychiatric co-morbidities such as depression, which further worsen function and quality of life [2].** Depression in ASD is more difficult to treat than in the general population, and certain treatments for depression can actually worsen core symptoms of autism or cause significant behavioral side effects. A Cochrane review found limited evidence for the effectiveness of SSRIs in the treatment of depression in adults with Autism, and no efficacy in children [3]. Furthermore, there are no evidence-based biological treatments for the core symptoms of autism. **The development of new strategies to treat both the core symptoms of autism and depression in patients with Autism may therefore have major clinical implications.**

**Repetitive transcranial magnetic stimulation (rTMS) is a non-invasive brain stimulation technique which is currently FDA-approved as a treatment for major depressive disorder for treatment refractory depression in adults [4].** Several large, multi-center, randomized controlled trials have clearly demonstrated the efficacy of TMS to treat depression, with remission rates 4.2 times greater than sham devices [5]. There is also evidence that when combined with pharmacotherapy, the remission rate is much greater [6]. Continuation studies found strong durability of response, with 64-90% of patients maintaining benefit from rTMS at 3-12 months after end of treatment [4]. While multiple different rTMS protocols have been tested to treat depression, the most widely studied and current FDA recommended protocol is high frequency (10 Hz) rTMS at 120% of motor threshold (MT) on the left dorsolateral prefrontal cortex (DLPFC) for 3000 pulses per session; daily (weekday) sessions are recommended for four to six weeks [4].

**The use of TMS in ASD generally has begun to be studied only recently, and standardized protocols for treatment in a clinical setting have yet to be established, but there is evidence of clinical benefit and symptom reduction for core autism symptoms.** There has been significant variation in choice of specific TMS protocols, and there is no consensus on the superiority of any one particular protocol or parameter for treating the core symptoms of autism. A majority of the studies have focused on low frequency rTMS (0.5-1.0 Hz), and have found improvement in multiple clinical symptoms including irritability and repetitive behaviors, as well as reduction of error rates and normalization of EEG recordings during visual processing tasks [7-9]. There have also been studies that used high frequency rTMS (ranging 5-8 Hz) which found significant reduction in social relating symptoms and self-oriented anxiety during difficult and emotional situations [10] and improved hand-eye coordination in individuals with low-functioning autism [11].

**While there have been single case studies [12] and anecdotal reports of successfully using TMS specifically for the treatment of depression in patients with Autism, there have been no standardized trials specifically addressing its effectiveness.** As noted above, there have also been few studies which examine the effect of high frequency rTMS on core autism symptoms. Given the increased difficulty in treating depression in autism, including resistance to or intolerance of many of the standard medication therapies, the establishment of safe and effective alternatives is a major area of need in this population. The safety of high frequency rTMS in the general population has been well established. The average risk of seizure is <0.1% for each patient or <0.003% per session, and the overall risk for seizure is slightly lower than that for antidepressant medications [4]. Additionally, in the current body of work using TMS to treat autism, no seizures have been induced in any trial, including those where high frequency

was used [13]. By using a protocol with widely established efficacy and safety, risk is minimized while allowing a better understanding of both treatment of depression in autism and the potential benefit of high frequency stimulation on core autism symptoms and deficits.

**The goal of this proposal is to investigate whether a standard rTMS protocol for depression, including multiple sessions applied to left dorsolateral prefrontal cortex (DLPFC) results in reduction of depressive symptoms for adult patients with ASD and MDD (Aim 1). The secondary goal is to investigate and whether there is any beneficial reduction in the core symptoms of autism (Aim 2).** There are two additional exploratory sub aims, for which the goal is to evaluate durability of response (Aim 1 and 2) and to evaluate for normalization of altered patterns of brain connectivity during cognitive processing tasks (Aim 2). These aims will be examined through an open-label study in which 10 participants will receive 25 sessions of rTMS at 10 Hz on the left DLPFC via daily (weekday) treatments (1 session per day, 5 days a week) over the course of 5 weeks.

**Aim 1. Determine the safety and therapeutic efficacy of left-sided DLPFC high frequency rTMS on MDD symptoms in patients with ASD:** We hypothesize that patients receiving the rTMS will tolerate the treatment course without difficulty and have clinically significant reduction of depressive symptoms after receiving all 25 sessions, as compared with their symptom burden prior to initiating TMS. Depression symptom data will be collected as pre- and post-TMS scores on Hamilton Depression Rating Scale (HAM-D). Depression scores will also be monitored periodically during course of TMS with Patient Health Questionnaires (PHQ-9).

**Exploratory sub-aim - Monitoring for durability of response:** We hypothesize that subjects receiving rTMS will durability of response in their depression symptom reduction, as measured by HAM-D scores at 1 month and 3 months post-TMS.

**Aim 2. Determine the effect of left DLPFC rTMS on core symptoms of ASD:** We hypothesize that subjects will experience reduction in core symptoms of ASD after completing all 25 sessions, as compared with their symptom burden prior to initiating treatment. For social and communication deficits, informant and/or self-report evaluations will be made pre- and post-TMS with the Social Responsiveness Scale (SRS), the Ritvo Autism Aspergers Diagnostic Scale-Revised (RAADS-R) and the Aberrant Behavior Checklist (ABC). Repetitive and restricted behavior will be evaluated using the Repetitive Behavior Scale-Revised (RBS-R), the ABC, and RAADS.

**Exploratory sub-aim: Determine if there are changes in performance during face and object processing tasks in patients with Autism who receive rTMS:** We hypothesize that there will be improvement from baseline scores in performance on behavioral processing tasks.

**Exploratory sub-aim - Monitoring for durability of response:** We hypothesize that subjects receiving rTMS will exhibit durability of response in their ASD symptom reduction, as measured by ABC, SRS, RAADS, AND RBR scores at 1 month and 3 months post-TMS.

## 2.0 Background

**Autism Spectrum Disorder (ASD) is a common and chronic disorder in the United States, which can cause significant limitations in ability to function and quality of life for the individual, and results in major costs to society in general.** Estimated current prevalence of ASD in the general population is between 1-2%, and has increased significantly in the past decade [14]. Per CDC reporting, prevalence per 1000 children in the United States increased from 6.7 in 2000 to 14.6 in 2012[15]. The total cost of ASD in the U.S., including medical costs and loss of productivity, is estimated at \$236 to 262 billion annually, with the majority of cost burden due to adult services, estimated \$175-196 billion [16]. Despite the enormous need for autism services, there are no evidence-based biological treatments for core ASD symptoms. The two medications with FDA-approval for patient with ASD, risperidone and aripiprazole, help mitigate associated symptoms such as aggression or irritability. Additionally, patients with ASD frequently have at least one, and often multiple comorbidities, including depression, with the estimated overall rate of comorbid psychiatric disorders in adults with ASD equal to 54% [17]. Although the prevalence of major depressive disorder (MDD) in patients with ASD has been hard to establish due to the difficulty of diagnosis in the setting of autism, estimates for prevalence of MDD in children are at least 10-14% [18] and 26% in adults [17], significantly higher than the general population. When depression is present as a comorbidity in ASD, patients report higher number of symptoms than those without ASD [19] and are at increased risk for suicidal ideation and suicide attempt than neurotypical individuals with depression [20]. In addition to the increased burden of depression on patients with ASD, there is limited data showing efficacy of standard pharmacological treatment for depression in both adult and pediatric patients with autism [3]. Furthermore, standard pharmacological therapies for depression have no benefit for the core symptoms of ASD, and can sometimes exacerbate them [3, 21].

**Repetitive transcranial magnetic stimulation is a non-invasive brain stimulation technique that is able to alter cortical excitability via the use of magnetic fields. It is an FDA-approved treatment for depression, and is being investigated as a potential treatment modality for a wide range of other psychiatric and neurological illnesses, including autism.** TMS works through a pulse generator, which transmits energy in the form of magnetic fields through a stimulating coil that is placed on the scalp. These fields are then transduced back into an electrical current by cortical neurons, specifically by neuronal axons (as opposed to the cell body itself). In rTMS, the magnetic pulses are biphasic, and pulsed at varying frequencies which can alter the pattern or type of cortical stimulation. High frequency (fast) rTMS is defined as greater than or equal to a frequency of 5 Hz and is generally considered to be excitatory stimulation, while low frequency (slow) rTMS is between 0.5-1 Hz and is considered to be inhibitory stimulation. Other factors which can alter the effect of the stimulation include the placement site and angle of the TMS coil, the configuration of the coil itself, the power of the stimulation (generally expressed as a percentage of motor threshold), the timing of the pulses, the number of pulses over a single session and total pulses delivered over all sessions.

Direct stimulation via rTMS occurs only within the first few centimeters of cerebral cortex. However, due to the immense connectivity within the brain, often to distant regions and across hemispheres, altered patterns of activity can be observed in multiple brain regions after stimulation at single location [22, 23]. Stimulation sites are chosen based on their presumed role in the targeted illness, and also on the anatomy of the head, as some locations do not allow for adequate penetration of the stimulus due to skull and brain structure. The dorsolateral prefrontal cortex is almost universally chosen as the stimulation site for depression due to both being an ideal site for rTMS (short scalp to cortex distance) and its known importance in executive functioning, motivation, and memory, as well as its high connectivity to other areas more directly related to emotional symptoms such as the ventromedial prefrontal cortex and the limbic system. High frequency (excitatory) rTMS left DLPFC is the most well-established protocol to

date, and is based on findings that show hypo-activity in the DLPFC along with measurable atrophy in depressed patients [24, 25].

**Autism Spectrum Disorder is widely theorized to involve abnormal balance of excitatory and inhibitory signaling and altered functional connectivity within and between different brain regions.**

These abnormalities in signaling and connection are thought to be the cause of both social and non-social core symptoms of autism, which reflect a mixed state of inappropriate signals and dysfunctional connections, rather than due purely to a deficit in either inhibitory or excitatory signaling. Specifically, there is a growing body of evidence that autism results in patterns of local overconnectivity and global or long distance hypoconnectivity [26]. In particular, there is a lack of functional connectivity between different brain regions, in which these distant regions function together in higher level processing of stimulus input and regulation/modulation of stimulus response (i.e. top-down processing) [27]. This has been shown to contribute to a vast array of deficits seen in autism, including visual imagery and language [28], working memory [29], social and emotional tasks [30], problem solving [31], response inhibition [32], and theory of mind [33].

**The focus of existing studies using TMS to investigate ASD has been wide-ranging, from the use of TMS to further describe and understand the underlying neuropathology and circuitry of ASD, use as a diagnostic tool, and as a new treatment modality with multiple different target symptoms.** The TMS protocols in these studies have also varied significantly, from number and type of pulses (single pulse, paired pulse, rTMS, theta burst), pulse frequency (Hz), length and number of exposures/treatments, and location of stimuli. A significant amount of early focus on rTMS specifically as a potential therapeutic tool has focused on loss of lateral inhibition and plasticity in individuals with ASD. Based on extensive histoanatomical, electrophysiological and other studies, some theories suggest that development of the functional units of the cortex, the minicolumn, is altered in autism, due to abnormal migration patterns of inhibitory interneurons and excitatory neuroblasts, which will eventually become pyramidal cells [34]. Briefly, the minicolumn is composed of centrally located pyramidal cells which form dyads with peripherally located interneurons. The abnormal migration of neurons results in loss of the inhibitory neurons in the periphery (peripheral neuropil space) as well as smaller pyramidal cell bodies, which are both reflected as an overall narrowing of minicolumns. The loss of interneurons leads to impaired lateral or surround inhibition, creating both a hyperexcitable state within the cortex, as well as a loss of appropriate directionality for excitatory stimuli. The decrease in pyramidal cell bodies creates a predisposition to shorter corticocortical connections, resulting in impaired plasticity and further predisposition to local hyperactivity.

The particular protocol developed by Casanova et al (2003) using low frequency, subthreshold rTMS to dorsolateral prefrontal cortex (DLPFC – either left sided or sequential bilateral) has focused on improved lateral cortical inhibition, based on the above theories regarding altered minicolumn development. Multiple studies using this protocol have found benefits in several different domains including improvement in irritability and repetitive behaviors and enhanced autonomic balance [8, 35]. They also found that gamma bands, as measured by EEG and associated with attentional processing and object processing, were significantly normalized when subjects were presented with a task requiring differentiation between illusory and non-illusory objects [7, 9], as well as having improving rates of error detection and normalization of event-related potentials (another EEG measurement which evaluates early and late stage processing of visual stimuli) [36, 37]. However, it is also relevant to note that none of these studies saw improvement in social functioning symptoms on any of the clinical scales. Additionally, some of the ERP data appeared at times contradictory between studies, and it was unclear what the relevance, if any, was to some of the changes measured.

Other studies have focused more explicitly on addressing the social functioning and communication deficits that are present in ASD, and have used both high frequency and low frequency TMS. Multiple, sometimes overlapping, theories have been proposed to explain the social symptoms in ASD, including theory of mind, broken mirror theory, and deficits in social motivation. The broken mirror theory suggests that deficits in the mirror neuron system (MNS), which consists of brain regions that are activated both when observing an action and performing that action, contribute to difficulty with social imitation and theory of mind [38, 39]. However, a recent comprehensive review found there is mixed evidence supporting this theory [40], and a TMS study evaluating interpersonal motor resonance (IMR, a measure of MNS function) had conflicting data regarding if and how a defect in MNS impacted social functioning[41].

“Theory of Mind” (ToM) relates to the ability to attribute mental states (e.g. beliefs, intents, desires, pretending, knowledge) to oneself and others and to understand that others have beliefs, desires, intentions, and perspectives that are different from one's own. Mentalizing is an aspect of ToM, which refers to the the ability to perceive and interpret human behavior in terms of intentional mental states. Multiple studies have found deficits in measures of ToM function in the autism population, specifically in terms of ability to understand and predict the mental states of others rather than in one’s self [42]. Additionally, areas of the prefrontal cortex have been routinely implicated as vital to processing mental states and theory of mind tasks, including the dorsolateral prefrontal cortex [43, 44], the ventromedial PFC [45], and the dorsomedial PFC [46, 47]. While the DMPFC and VMPFC appear to play a greater role in theory of mind and social cognition, the DLPFC also appears to play a significant but potentially more indirect role.

All of this data is congruent with a recently developed theory that speculates that deficits in social and communication domains in autism may be largely due to a failure of top-down processing (i.e. complex processing of stimuli that modulates and regulates how we respond to the stimuli) [48]. A high frequency (5Hz) rTMS protocol was used in one study based on these theories of dysregulation of top-down processing, specifically using an H-type coil which allows deeper penetration (often called deep TMS) of the magnetic field into the cortex, in attempt to target the bilateral dorsomedial PFC. After 10 sessions, patients showed significant improvement on the Social Relatedness Subscale of the Ritvo Autism Asperger Diagnostic Scale, but not on other ASD scales (e.g. Autism Spectrum Quotient) or experimental measures of mentalizing [10]. Of note, the study used the lowest possible frequency that could still be considered “high frequency” and may also have been limited by the smaller number of sessions. Additionally, by nature of the H coil design which generates fields with much greater surface area, multiple areas other than the dmPFC could have also received at least partial direct stimulation.

As discussed earlier, there is strong evidence for significant abnormalities in global functional connectivity and excitatory/inhibitory imbalance and their likely role in driving many core symptoms of autism. Repetitive transcranial magnetic stimulation is known to cause changes in both local and distant patterns of brain activity, and has shown normalization of hypoactive functional pathways. Given this, **we hypothesize that in addition to treating symptoms of depression, rTMS may also result in reduced symptoms of autism due to improved functional connectivity.**

**Rationale for study design:** Significant consideration was given to the question of which TMS protocol parameters to use. Our primary aim was to establish efficacy of rTMS to treat depression in patients with Autism Spectrum Disorder. In order to accomplish this aim and minimize risk to the participants, a standard, previously established protocol was chosen with FDA approval and established efficacy and safety in the general population. While there are other protocols which have found similar efficacy in using low frequency rTMS to treat depression, these protocols remain investigational in nature and are not yet FDA approved. There is theoretically slightly higher risk of seizures with high frequency rTMS as

opposed to low frequency rTMS, however, this risk is extremely low overall and, and there was overall greater justification for choosing an FDA approved protocol which used high frequency stimulation for depression rather than an investigational protocol that used low frequency stimulation. Additionally, our theorized goal in targeting ASD symptoms is to improve functional connectivity along hypoactive pathways, and high frequency stimulation will likely be more effective at achieving this as it is excitatory. This goal will be indirectly investigated via comparison of pre and post-TMS testing of cognitive processing of visual social and non-social stimuli through behavioral tasks.

### **3.0 Intervention to be studied**

Repetitive transcranial magnetic stimulation (rTMS) is a non-invasive brain stimulation technique which is currently FDA-approved as a treatment for major depressive disorder for treatment refractory depression [4]. Several large, multi-center, randomized controlled trials have clearly demonstrated the efficacy of TMS to treat depression, with remission rates 4.2 times greater than sham devices [5]. There is also evidence that when combined with pharmacotherapy, the remission rate is much greater [6]. Continuation studies found strong durability of response, with 64-90% of patients maintaining benefit from rTMS at 3-12 months after end of treatment [4]. While multiple different rTMS protocols have been tested to treat depression, the most widely studied and current FDA-recommended protocol is high frequency (10 Hz) rTMS at 120% of motor threshold (MT) on the left dorsolateral prefrontal cortex (DLPFC) for 3000 pulses per session; daily (weekday) sessions are recommended for four to six weeks[4].

No medications are being investigated as part of this study beyond current participant medication regimen being recorded. There is no control or placebo group, data will only be compared within the same participant group on a pre- and post-intervention basis. For further information regarding the safety of TMS, please see sections 2 and 15.

### **4.0 Study Endpoints (if applicable)**

Primary study endpoints:

- Statistically and/or clinically significant reduction in symptoms of MDD after completion of TMS protocol
- Statistically and/or clinically significant reduction in core symptoms of ASD after completion of TMS protocol

Secondary study endpoints:

- Measurable and statistically significant change in neurological patterns of activity after completion of TMS protocol
- Determine latency and durability of symptom reduction effect at 1 and 3 months after completion of TMS protocol

Primary safety endpoints:

- Prolonged or significant worsening of MDD symptoms, including any occurrence of suicidal ideation, during the course to TMS protocol
- Prolonged or significant worsening of ASD symptoms during the course of the TMS protocol

- Seizure

Secondary safety endpoints:

- Psychological or medical inability to undergo TMS or other study component

## **5.0 Inclusion and Exclusion Criteria/ Study Population**

### **Inclusion Criteria:**

- Age range: 18-65 years old
- Patients must meet DSM-V criteria for both Autism Spectrum Disorder (ASD) and Major Depressive Disorder (MDD) as determined by formal clinical psychiatric interview and supported by prior diagnostic evaluation when available
- Patients on pharmacological therapy for MDD or ASD symptoms prior to initial assessment will included, but must be stabilized on regimen at least one month prior to enrollment, and will be continued on these therapies throughout the study
- Subjects with mild intellectual disability (ID) as defined by a full-scale IQ between 60 and 70 will not be excluded from the study if they meet all inclusion criteria. ID is a very common comorbidity in patients with autism, with a 45% rate of incidence [49]. To exclude subjects with any level of ID may result in significant limitations to both recruitment and generalizability of the results. Subjects without prior formal IQ testing will be evaluated for level of disability via the Wechsler Abbreviated Scale of Intelligence-Modified during initial assessment. If the patient has a full-scale IQ between 60 and 70, informed consent will be obtained from the subject's legally authorized representative.
- Patient's currently taking bupropion (Wellbutrin) will not be excluded from the study, provided they are taking a fixed and stable dose of the medication.

### **Exclusion Criteria:**

- Uncontrolled and/or untreated seizure disorder as defined by any incidence of seizure within the past 6 months. Patients with diagnosed epilepsy, or prior seizures, will be allowed in the study if they are taking an anticonvulsant medication, or have not had a seizure in the past year off medications.
- Moderate to severe intellectual disability (ID) as defined by IQ < 60, determined by prior IQ testing or Wechsler Abbreviated Scale of Intelligence (WASC-II) if no prior test results available
- Other psychiatric or neurodevelopmental illness that is the primary area of clinical focus (including but not limited to primary psychotic disorder, substance abuse disorder, and ASD or ID which are secondary to genetic syndromes)
- Active suicidal ideation or suicide attempt in the 90 days prior to initial assessment
- Presence of any metal implants or devices in the head or neck (e.g. metal plates or screws)
- No participants who are pregnant or who are planning to become pregnant



- Have surgical implants such as pacemakers or cochlear implants
  - Have ever suffered a closed head injury or concussion
  - Are currently under the influence of alcohol or other recreational drugs
  - Cannot understand task instructions
  - Are currently enrolled in a course in which the PI or co-Is are instructors
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- Inability or unwillingness of participant or legal guardian/representative to give informed consent
  - There will be no discrimination or exclusions based on race, gender, sexual orientation, or other socioeconomic factors. Of note, while both male and female participants will be actively and equally recruited using the same methods. The natural distribution of autism in the population skews towards significant towards male gender, with male prevalence being 4-5 times that of female prevalence. Our study will therefore likely have more male participants than female due to this trend in prevalence.
  - Children (age <18) are being excluded from this study for several reasons. While autism is a pediatric neurodevelopmental disorder with symptom onset as young as one year of age, it is also one that is chronic throughout adulthood. Both children with autism and neurotypical children undergo periods of rapid change in brain size, structure, and organization as they age, and the interaction between a full rTMS series and brains that are still involved in periods of very active development and whom may also be at different points along their own developmental timelines may skew or alter the data that is collected. Additionally, due to both brain growth and increases in skull thickness, children of different ages may have significantly different “scalp to cortex” distances, which can result in very different patterns of cortical stimulation despite uniform coil positioning. This will be an added, unnecessary variable which would compromise the attempt at performing a standardized protocol. Finally, while high frequency rTMS is an FDA approved treatment for depression in adults, it has not yet been FDA approved in children and adolescents.

## **6.0 Number of Subjects**

The total number of subjects to be recruited is 15. The trial is open-label and consists of only one treatment modality, and therefore does not involve any randomization.

## 7.0 Setting

**Clinical scales and TMS procedure.** All rTMS sessions will take place at the MUSC Institute of Psychiatry, in the Brain Stimulation clinic and research suite on the 5<sup>th</sup> floor. The same Neuronetics/Neotonus TMS device will be used for all procedures. The device is maintained by the Brain Stimulation Department and is kept in a locked room when not in use. Clinical scales will be given either at the Institute of Psychiatry or at 30 Bee Street in a private room. Video and music will be made available to patients prior to and during treatment to help reduce stress. Patients will also be allowed to bring video games or other distractor or “comfort” items with them to each treatment session, provided they do not interfere with the function of the TMS device.

## 8.0 Recruitment Methods

Recruitment will occur on an ongoing/rolling basis over one year after final project approvals and finalized budget. Potential subjects will be recruited through a variety of online avenues and local autism community groups and organizations, as well as advertising to the general public.

- Project Rex is an autism-focused group run by Dr. Gwynette at MUSC, and has a robust social media network as well as a large patient directory. Information regarding the study and digital copies of fliers will be posted on social media forums (Facebook, Twitter, Instagram) under existing Project Rex accounts, as well as sent via email to the Project Rex mailing directory. While the Project Rex patient directory does not contain specific information regarding comorbid depression, Dr. Gwynette may discuss the project with patients he is actively treating as well as their family members. The study may also be advertised at any other groups, projects, or events that are being run by Project Rex.
- In addition to Project Rex, investigators will reach out to other local autism organizations and groups such as the Lowcountry Autism Foundation (LAF), the Lowcountry Autism Consortium, South Carolina Autism Society, and Charleston Young Adult ASDs to provide information regarding the study to their members, as well as digital and/or physical copies of fliers to distribute to their members if they are willing to do so. Investigators will also offer come to speak about the study and answer questions during group meetings in person for any groups who are interested. Emails will be distributed by the investigators to families and stakeholders on the Project Rex newsletter distribution list and the MUSC broadcast email system. The study investigators will also distribute flyers to families and stakeholders at the 2016 Lowcountry Autism Forum, held at the College of Charleston on October 15, 2016.
- The investigators will also mail study flyers to MUSC patients with a previous diagnosis of Autism Spectrum Disorder, as identified using MUSC Epic's query technology. In addition, the investigators will reach out to MUSC physicians treating patients meeting enrollment criteria based on MUSC' Epic's query capability.
- Investigators will reach out to local colleges and universities, including College of Charleston, Trident Technical University, and Charleston Southern University and provide them with information regarding the study and request permission to post physical copies of fliers in high

traffic areas on campus. Fliers will also be posted around the MUSC campus and hospital once approved.

- Investigators will reach out local psychiatry offices and provide them with information regarding the study and digital and/or physical copies of fliers for them to share with their patients at their discretion.
- Flyers and posters will contain information including a basic explanation of study (investigating the use of non-invasive brain stimulation for the treatment of depression in patients with autism), eligibility (age range, diagnosis of MDD and ASD), and information about financial compensation and ability to receive FDA approved treatment for depression at no cost to patient. They will also include study site and contact information for study coordinator(s).

## **9.0 Consent Process**

- The subjects will be consented in person by one of the study investigators.
- The consent process will take place in the MUSC Department of Psychiatry Brain Stimulation Clinic.
- The investigator will read over the consent form alongside the subject, and will be available to answer any questions that the subject has in the moment.
- There will be no waiting period between informing the prospective subject and obtaining the consent
- Spanish interpreters and translated informed consent documents and HIPPA information documents will be available for Spanish-speaking participants.

### ***For Cognitively Impaired Adults***

- Subjects with a full-scale IQ less than 70 will require the participant's parent or legal guardian to provide informed consent for participation in this study.
- In the case of intellectually impaired individuals, the parent or legal guardian will help interpret whether the child understands and agrees to participate.
- Throughout each visit to the lab, the experimenters allow the participant to indicate that they need breaks or that they do not want to participate any longer.

## **10.0 Study Design / Methods**

### **Figure 1.**

**General overview:** Our primary aims are to determine the safety and efficacy of 25 sessions of 10 Hz rTMS to the left DLPFC as a potential therapy for treatment of depression in adults with ASD (Aim 1) and for treatment of core autism symptoms (Aim 2). The aims of the study will be accomplished by performing an open-label pilot study with 10 patients who meet diagnostic criteria for MDD and ASD. Participants will be recruited from the local community and specifically from local autism groups (including Project Rex) and from MUSC outpatient psychiatric clinics. The majority of this study will take place over the five weeks of rTMS treatments during which both of the primary aims will be accomplished. We will also collect data at 1-month and 3-months post treatment to determine the durability of the response to treatment.

Initial Assessment	<ul style="list-style-type: none"> <li>Structured Clinical Interview</li> <li>Pre-TMS clinical scale assessments</li> <li>IQ testing</li> <li>Site tour</li> <li>fMRI training/mock scanning session</li> </ul>
Week 1	1) First session: <ul style="list-style-type: none"> <li>Pre-TMS task-based fMRI scan</li> <li>Motor threshold testing</li> </ul> 2) Daily <u>rTMS</u> treatment session (5x/week)
Weeks 2-4	Daily <u>rTMS</u> treatment session (5x/week)
Week 5	1) Daily <u>rTMS</u> treatment session (5x/week) 2) Final session: <ul style="list-style-type: none"> <li>Post-TMS clinical scale assessments</li> <li>Post-TMS fMRI task (within 2 weeks)</li> </ul>
Week 9	1 month follow up clinical scales
Week 17	3 month follow up clinical scales

**Initial assessment:** We will enroll, screen, and assess participants during an ongoing enrollment period. Any documentation available for each participant regarding prior diagnostic testing for ASD, MDD, or IQ will be requested from the participants themselves. Records may be requested by the investigators from outside clinics or facilities or accessed from MUSC records after explicit patient permission is granted via signed records release form. All copies of patient records obtained for this study will be destroyed (physical copies) or erased (electronic copies) once relevant data is recorded in a de-identified manner, and electronic data will be stored and transmitted only via secure MUSC servers. Each participant will be evaluated for psychiatric disorders including ASD and MDD via structured clinical interview based on DSM-V criteria. Participants without prior formal IQ testing will also undergo an IQ assessment. Female subjects will be given a pregnancy test. During the initial assessment, the participants will also be given a tour of the TMS facilities and device. Initial assessment will take between **1.5-3 hours**, depending on interview and testing length.

**Testing and treatment phase:** Prior to initiation of rTMS treatments, participants will be evaluated via standardized clinical scales with established validity for their baseline symptom burden for both depression and autism and scores will be recorded (see Table 1). The HAM-D will be used for participants both with and without intellectual disability, as its use in patients with mild to moderate ID is generally accepted and considered accurate [50]. For participants who have eligible informants, the ABC and RBS-R will be used and will be based on informant report; all participants will receive the RAADS and SRS evaluation. Participants will also perform cognitive processing tasks prior to rTMS to establish baseline scores. Clinical symptom scales will occur during the initial assessment, and the pre-TMS cognitive processing tasks will occur on the same day but prior to the first rTMS treatment. Participants will receive rTMS treatments five days a week for five weeks for a total of 25 sessions. Allowances will be made for missed appointments or holidays, in order for each person to receive 25 treatment sessions within 6 calendar weeks. During the course of the rTMS treatments, participants will be asked at each treatment session about any side effects they have experienced since the last treatment, as well as if they have experienced any change in ASD or MDD symptoms. The PHQ-9 will be administered twice a week for the duration of the rTMS therapy, for additional monitoring of MDD symptoms.

**Post-treatment/continuation phase:** After the last rTMS treatment, each participant will be evaluated using the same series of standardized clinical scales he or she initially took prior to receiving rTMS, to evaluate any change to symptom burden (with the exception of IQ testing). The post-treatment and follow-up RAADS will be modified slightly from the pre-treatment RAADS by changing the possible answer options from “*True now and when I was young*” / “*True only now*” / “*True only when I was younger than 16*” / “*Never true*” to “*True or almost always true*” / “*Never true or rarely true*”, with reverse coding as usual. This change is to allow for evaluation of any change in symptom burden, and is very similar to a modification made in another study (Enticott, 2014) evaluating the effect of TMS on ASD symptom burden.[10] Participants will also repeat the cognitive processing tasks at this time to establish post-TMS scores. The clinical scales will also be repeated at 1-month and 3-months after the last rTMS treatment to monitor for durability of response. For participant’s convenience, investigators will offer to coordinate durability evaluations with any existing appointments at MUSC which are unrelated to this study, for the participant’s convenience.

**Clinical scales:** Clinical scales will be performed per the schedule in Table 1. All clinical scales in every phase will be performed by one of the investigators or research assistants who has completed the recommended training in how to administer and score these evaluations. All scales are used in their standard format except the post-treatment and follow-up RAADS as discussed above. Each separate evaluation takes between 5 to 30 minutes for the participant. Pre-TMS evaluation time is included as part of overall initial assessment time. Total required time for post-TMS clinical scales will vary depending on participant, but will take approximately **20 minutes to 1 hour** for each time point (immediate post, 1-month post, 3-month post).

**Cognitive processing tasks:** This portion of the study will occur in three separate phases:

- 1) Training session to introduce the type of tasks that will be performed.
- 2) Participants will perform pre-TMS cognitive processing tasks, with performance scores and reaction times being recorded. This will occur on the same day as the initial rTMS treatment session, prior to receiving rTMS.
- 3) Participants will perform the same cognitive processing tasks after the final rTMS treatment, either at the same session as the last treatment or during a separate session within two weeks after the last session.

**Training session:** Participants will be introduced to the two types of tasks they will be performing (see below) and allowed to practice several times before making the real attempt. This will take approximately 15-20 minutes

**fMRI tasks and scanning:** Subjects will complete two tasks on a laptop running eprime software: (a) a dot-probe task and (b) task-switching.

For the dot-probe task, two visual stimuli are presented simultaneously then removed, with the location of one image replaced by a probe (Posner, 1980). Participants respond with the left or right hand to indicate the probe location. In this type of probe task, brain activation is expected to be higher for probes that are in the same location as the stimulus that is attended more. Reaction time is also expected to be faster if the probe appears in a location that was just recently attended compared to the location that was not attended. In one condition, the stimuli are a face and house to assess attentional bias to faces versus non-faces (related to social awareness). Another condition presents a face with direct gaze and one with averted gaze (related to social motivation, social anxiety). These two conditions are implemented in two functional runs lasting 5 minutes each. The runs are presented in counterbalanced order across subjects.

**Cued task-switching:** This task is modeled after Gruber et al (2006), but with some modifications. In an event-related design, participants categorize a stimulus according to two dimensions, shape or color. Two abstract shapes and two colors are used to create four unique stimuli. On a given trial, the subject is cued to categorize the object (shape 1 requires a right index finger button press; shape 2 requires a right middle finger button press) or the color (red requires index and blue requires middle finger button press). The cue is a square to indicate an object categorization or a diamond to indicate a color categorization. Congruent trials are those in which the relevant and irrelevant dimensions map onto the same response; incongruent trials are those in which the dimensions map onto different responses. A task switch trial is defined as switching from the object to the color task relative to the previous trial. The cue appears for 500 ms prior to the target (with cue onset jittered 0, 100, or 350 ms relative to volume collection; note that Gruber et al. varied preparation time but we keep this value constant in the present study as our goal is not to separate out preparation from response phases). The cue remains on the screen while the target is displayed for 750 ms, followed by a response period of 900 ms, followed by a filler blank screen for a variable duration so that the entire trial duration is 2.5 sec (or one TR). We opt to keep the response window the same on all trials as a longer jitter time leaves a shorter response window than shorter jitter times, potentially leading to more missed responses on those trials. A single run consists of 128 trials, with the eight stimulus (4) x task (2) combinations equally distributed (and repeated 16 times per combination, yielding an equal number of congruent / incongruent trials. Trials are pseudorandomly ordered with approximately equal numbers of switch and repeat trials.

The tasks should take approximately 20-30 minutes to complete.

**rTMS protocol:** rTMS will be delivered via a Neuronetics/Neotonus Neopulse rTMS device with either a Neuronetics or Neopulse solid-coil head coil. Both coils are butterfly coils, and the magnetic firing properties of both are identical when measured by an oscilloscope. We will use a standard resting motor threshold (rMT) determination to determine the TMS dose[51]. Treatment will be delivered at a goal of 120% MT. If 120% of MT is not tolerable, treatments will be delivered at a minimum of 100% MT. During the first session, and during the first week, treatment will be allowed to be less than the ideal, down to 80% MT if needed. Each active rTMS treatment will consist of a total of 3000 pulses of 10Hz stimulation (4s-on, 10s-off) per treatment. Treatments will be delivered at the EEG coordinate for F3 (which approximates the left DLPFC), and will be found using the Beam-F3 method [52]. Each treatment will last 15 minutes, with the exception of initial treatment which will include obtaining the motor threshold and will require an additional 10-15 minutes. This is a treatment paradigm that has been studied extensively for depression and has been FDA approved (see: section 2 and 3). All motor threshold testing and initial F3 site location will be performed by co-investigator Dr. Greg Sahlem. Subsequent treatment will be performed by investigators or assistants with clinical training (MD or RN) who have received specific training in performing rTMS. The treatment provider will remain in the room for the duration of the treatment, and for another 10 minutes after treatment concludes to ensure participant safety and observation of any side effects or adverse events. Participants will be allowed to watch videos, play handheld games, or listen to music of their choice to help alleviate any stress or discomfort experienced during the treatment.

**Payment to participants:** In return for their time, effort and travel expenses, participants will be paid via a \$20 gift card at the completion of each of the twenty-five TMS treatments, for a total of \$500 for the complete course of TMS. In addition, participants will receive a \$50 gift card by mail for completing the outcome measures one month after treatment and a \$50 gift card by mail for completing the outcome measures three months after treatment. Therefore, the total amount of compensation for completing the entire study protocol will be \$600 in gift cards.

Table 1. Schedule of assessments.

	Assessment domain	Baseline	Post	1 and 3 month follow up
Screening and Enrollment Eligibility:				
Structured Clinical Interview for the DSM-V: Structured interview to determine Axis I psychiatric conditions based on DSM-5 criteria.	Psychiatric history	X		
Hamilton Rating Scale for Depression (HAM-D)[53]: Clinician administered measure of depressive symptoms.	Depressive symptoms	X	X	X
Wechsler Abbreviated Scale for Intelligence[54]: IQ testing for adults.	IQ	X		
Depression Outcomes:				
Hamilton Rating Scale for Depression (HAM-D) [53]: Clinician administered measure of depressive symptoms.	Depressive symptoms	X	X	X
Patient Health Questionnaire [55]: Brief clinician administered measure of depressive symptoms.	Depressive symptoms	X	Twice weekly during rTMS	
Autism Outcomes:				
Aberrant Behavior Checklist (ABC) [56]: Clinician administered rating scale assessing five problem areas: Irritability, Lethargy/Social Withdrawal, Stereotypy, Hyperactivity, and Inappropriate Speech, based on caregiver/informant reports.	Social functioning and repetitive/restricted behavioral symptoms	X	X	X
Social Responsiveness Scale-2 (SRS-2)[57]: Self-report rating scale assessing social interest and interaction.	Social functioning symptoms	X	X	X
Ritvo Autism-Aspergers Diagnostic Scale – Revised (RAADS-R) [58]: Self-report based assessment of autism core symptoms in adults including social relatedness, language and communication, and sensorimotor and stereotypies.	Social functioning and repetitive/restricted behavioral symptoms	X	X	X
Repetitive Behavior Scale – Revised (RBS-R)[59]: Caregiver/informant completed rating scale assessing repetitive and restricted behavior patterns.	Repetitive/restricted behavioral symptoms	X	X	X
Cognitive processing tasks:	Social and non-social cognitive processing;	X	X	

## 11.0 Specimen Collection and Banking (if applicable)

No specimens will be collected for this study.



## 12.0 Data Management

**Statistical Analysis:** Individual data in the form of scores from clinical symptom scales will be collected at the four specified evaluation time points (see Table 1). Both total scores and scores broken down by subscale will be analyzed when applicable. All data will be considered within-subject since all participants are receiving the same treatment and will be evaluated in the same way, and there is no control group or comparison groups receiving alternative therapy. This data will be analyzed using a one-way, repeated measures ANOVA test to examine differences between means, which is ideal for analyzing longitudinal (pre/post, multiple within subject repeated measures) data. Data will be assessed for distribution, variance and sphericity/compound symmetry prior to performing the ANOVA to ensure applicability of ANOVA to the dataset. Adjustments to analysis will be made as needed (according to accepted statistical guidelines) to correct for differences in variance or sphericity. Should data not fit normal distribution, non-parametric alternatives (such as the Friedman test) will be used. T-tests may also be used to directly compare score data between two specific time points.

**Sample Size Justification:** A total of  $n=15$  subjects will be recruited. We anticipate that 20 to 30% of study subjects will withdraw from the study or be lost to follow-up, meaning that  $\geq n=10$  subjects are expected to complete the study. This small sample size was chosen primarily based on being a pilot study to determine safety and potential efficacy. While the sample size limits the power of our study and increases the risk of a false rejection of our hypotheses regarding efficacy (Type II error), it will still provide enough relevant safety data upon which we can base future, larger studies in which efficacy can be better established. Additionally, the inclusion criteria of both ASD and MDD (in comparison with either ASD or MDD alone), as well as the nature of the intervention which requires participants to return to the study site frequently over several weeks, significantly limits recruitment potential. Therefore, a small sample size was also necessary to be able to meet recruitment requirements within a reasonable timeframe.

**Relapse, Drop-Out and Clinical Deterioration:** Every effort will be made to re-engage patients who miss appointments. Clinical deterioration, such as exacerbation of MDD or ASD symptoms, will be assessed on a case-by-case basis by the study physician and appropriate referral will be made throughout the treatment phase of this study, as well as via monitoring of PHQ-9 scores. Subjects will be considered drop-outs if they do not come back for follow-up visits after receiving two phone calls and two letters inviting them to return. With the exception of subjects who formally withdraw from the study, we will attempt to assess early terminators at the time of discontinuation and at the post-treatment time points. These subjects will be withdrawn from the study but considered in the intent-to-treat efficacy analyses.

**Strategies to Ensure a Robust and Unbiased Approach:** The proposed study will achieve robust and unbiased results via several design features including: explicit inclusion/exclusion criteria; use of validated laboratory and interview/self-report measures and methods; explicit hypotheses and corresponding planned statistical analyses; power estimates; planned handling of retention/attrition and missing data; and careful consideration of potential confounds. All experimental details are reported in a detailed and fully transparent manner to support replication.

**Data Security:** Research material obtained from individual participants includes questionnaires, task-based scores and responses, and interviews with study personnel. All of the non-task data will be directly input into REDCap which is a secure, password protected web-based data collection system. The only written research material with identifiers will be the informed consent and HIPAA documents. These paper records will be stored in an office in the Institute of Psychiatry that is locked when not in use.



All participant data will be recorded, transferred, and stored only via secured and encrypted MUSC networks. Within the secured REDCap network, participants' email, initials, and signature may be recorded in order to 1) allow some surveys to be sent electronically via email and allow REDCap to record those results and accurately assign them to the correct participant and 2) to accurately maintain compensation logs for each participant (initials and signatures). Other than the HIPAA and informed consent, any participant records or information regarding prior diagnosis that contains identifying information will be destroyed or erased after being recorded electronically in REDCap, and no identifiers will be entered into REDCap other than those mentioned above. Access to data will require input of current MUSC credentials and will be additionally password protected. Additionally, only our PI and co-investigators who are directly recording and entering participant data will be given access within REDCap to see identifiers, so any staff (current or future) who are not directly involved in working with participants will not be able to see participant's identifiable information. All investigators and assistants will have completed training in adherence to HIPAA requirements and will be certified via MUSC CITI training.

### **13.0 Provisions to Monitor the Data to Ensure the Safety of Subjects (if applicable)**

This section is based on the recommendations in NIDA's "Guidelines for Developing a Data and Safety Monitoring Plan" ([www.drugabuse.gov/funding/dsmbsop.html](http://www.drugabuse.gov/funding/dsmbsop.html)).

#### **A. Summary of the Protocol.**

This application proposes to investigate the effects of rTMS on depression symptoms in patients with Autism and the safety and efficacy of high frequency rTMS on core symptoms of autism. The primary outcomes of interest are depression symptoms (Aim 1) and core symptoms of autism (Aim 2). Inclusion/exclusion criteria are outlined above. Power calculations and sample sizes are in the Data Analysis Plan section.

#### **B. Trial Management.**

The study will be managed from the Brain Stimulation Laboratory Division and Community Outreach Division of the Department of Psychiatry and Behavioral Sciences at the Medical University of South Carolina. The target population is described in the inclusion/exclusion criteria detailed in Section 5.

#### **C. Data Management and Analysis.**

Data will be entered by research assistants directly into a computer using standard database software using REDCap. The data analysis plan is outlined in Section 10.

#### **D. Quality Assurance.**

Quarterly data audits will be conducted. Confidentiality protections are outlined above.

#### **E. Regulatory Issues.**

Potential conflicts of interest will be reported using the NIH rules for disclosure. Adverse Events (AEs)/Serious Adverse Events (SAEs) occurring during the course of the project will be collected, documented, and reported in accordance with protocol and IRB reporting requirements. All research staff involved with adverse event reporting will receive general and protocol specific AE/SAE training including identification, assessment and evaluation, and documentation and reporting. A research specialist will identify any potential adverse events during the course of the study from participant self-report and administration of the visit assessments and procedures. The research assistant will provide information to a study physician, who will be responsible for AE/SAE assessment and evaluation

including a determination of seriousness and study relatedness. Any significant actions taken by the local IRB and protocol changes will be relayed to NIDA.

#### F. Definition of AE and SAE.

An Adverse Event (AE) is defined as any untoward medical occurrence in a study subject administered a pharmaceutical product that does not necessarily have a causal relationship with this treatment (ICH GCP). Any unwanted change, physically, psychologically or behaviorally, that occurs in a study participant during the course of the trial is an adverse event. A Serious Adverse Event (SAE) is defined as an adverse event that has one of the following outcomes:

- Results in death,
- Is life-threatening,
- Requires inpatient hospitalization or prolongation of existing hospitalization,
- Results in persistent or significant disability/incapacity,
- Is a congenital anomaly/birth defect OR
- Requires intervention to prevent one of the above outcomes.

#### G. Documentation and Reporting.

AEs/SAEs are documented and reported as per protocol and IRB requirements. Research staff will identify adverse events and obtain all available information to assess severity, seriousness, study relatedness, expectedness, outcome and the need for change or discontinuation in the study intervention. Adverse events are generally documented on AE Logs and AE Case Report Forms (CRFs). Additional relevant AE information if available should be documented in a progress note in the research record as appropriate to allow monitoring and evaluating of the AE. If the AE meets the definition for serious, appropriate SAE protocol-specific reporting forms are completed and disseminated to the appropriate persons and within the designated timeframes as indicated above. For each AE/SAE recorded, the research staff will follow the AE/SAE until resolution, stabilization or until the participant is no longer in the study as stated in the protocol. When a reportable SAE is identified, the research staff will notify the MUSC Institutional Review Board (IRB) within 24 hours and complete the AE report form in conjunction with the PI. The MUSC IRB meets monthly and is located at 165 Cannon Street, Rm. 501, Charleston, SC 29425. Communication with the IRB is through email, memos, official IRB forms, and online reporting. A report will also be sent to the NIH program officer assigned to the project.

If complete information is not available when the initial 24-hour SAE report is disseminated, follow-up information will be gathered to enable a complete assessment and outcome of the event. This information may include hospital discharge records, autopsy reports, clinic records, etc. The research staff will attach copies of source documents to the SAE report for review by the PI and for forwarding to the NIH program officer as appropriate within 2 weeks of the initial SAE report. In addition, the PI will provide a signed, dated SAE summary report, which will be sent to the NIDA Medical Safety Officer within two weeks of the initial SAE report.

We will report adverse events to the Medical University of South Carolina (MUSC) Institutional Review Board (IRB) online as soon as possible, but no later than 10 working days after the investigator first learns of the event. The MUSC IRB AE reporting requirements are as follows: All deaths that occur during the study or 30 days post termination from the study are required to be reported as adverse events even if they are expected or unrelated. Other adverse events are reportable to the MUSC IRB if the AE is unexpected AND related or possibly related AND serious or more prevalent than expected. All three criteria must be met for an AE to be reported to the MUSC IRB. The IRB definition of unexpected is that the AE is not identified in nature, severity or frequency in the current protocol, informed consent, investigator brochure or with other current risk information. The definition of related is that there is a reasonable possibility that

the adverse event may have been caused by the drug, device or intervention. Reportable AEs are reviewed by the IRB Chair and reported to the IRB Board at the next meeting.

#### H. Trial Safety.

The potential risks and benefits and methods to minimize these risks are outlined above. The research staff will report any unexpected AEs or any scores of “severe” on the side-effect symptom rating form or any FDA-defined serious AEs to the PI within 24 hours so that the PI can decide on the appropriate action. All unexpected AEs will be monitored while they are active to determine if treatment is needed. Study procedures will follow the FDA’s Good Clinical Practice Guidelines ([www.fda.gov/oc/gcp](http://www.fda.gov/oc/gcp)). Any outside requests for information or any breaches in confidentiality will be reported to Dr. Gwynette.

An interim analysis is not planned at this time.

#### J. DSM Plan Administration.

Dr. Gwynette and Dr. Sahlem will be responsible for monitoring the study, and at least one of them will participate in weekly study meetings. A DSM report will be filed with the IRB and NIDA on a yearly basis, unless greater than expected problems occur. The report will include participant characteristics, retention and disposition of study participants, quality assurance issues and reports of AEs, significant/unexpected AEs and serious AEs. We will report outcomes at the end of the trial.

#### J. DSM Board.

A Data Safety and Monitoring Board will be formed to monitor both the rate and severity of adverse events. This panel will include clinicians with expertise in autism and depression and a statistician.

#### K. Risk Benefit Ratio.

The assessments and questionnaires are non-invasive and have inherently minimal risks. Potential risks of concern are loss of confidentiality and adverse events to rTMS. As discussed in sections 10, 14, and 15, our research team will attempt to minimize these risks. Knowledge gained by the proposed study would help fill an important void in development of a potential treatment for depression in autistic patients, as well as a potential treatment for autism itself.

### CLINICALTRIALS.GOV REQUIREMENTS

In accordance with Public Law 110-85, this project will be registered at the ClinicalTrials.gov Protocol Registration System Information Website prior to study initiation.

## 14.0 Withdrawal of Subjects (if applicable)

**1) Drop-outs:** Every effort will be made to re-engage patients who miss appointments. Subjects will be considered drop-outs if they do not come back for follow-up visits after receiving two phone calls and two letters inviting them to return.

**2) Clinical deterioration and safety concerns:** Clinical deterioration, such as exacerbation of MDD or ASD symptoms, will be assessed on a case-by-case basis by the study physician as well as via monitoring of PHQ-9 scores, and appropriate referral will be made throughout the treatment phase of this study. Any patient reporting suicidal ideation or severe worsening of symptoms will be immediately withdrawn from further rTMS treatments. Any subject who experiences a seizure during the course of the treatment phase will be immediately withdrawn from any further rTMS treatments. If subjects experience significant

claustrophobia or panic during any portion of the fMRI paradigm or during any of the rTMS treatments, they will be withdrawn or partially withdrawn from the study.

**3) Partial withdrawal:** With the exception of subjects who formally withdraw from the study, we will attempt to assess early terminators at the time of discontinuation and at the post-treatment time points. These subjects will be withdrawn from the study but considered in the intent-to-treat efficacy analyses. Participants who cannot tolerate fMRI paradigm will still be allowed to participate in the rTMS treatment and clinical scale evaluation.

## 15.0 Risks to Subjects

**Potential risks of rTMS:** The use of high frequency rTMS has been FDA-approved for the treatment of major depressive disorder since 2008. Our stimulation parameters are identical to the FDA approved protocol (3000 pulses, 10Hz, 4-Seconds On, 8-Seconds off), and has been used safely in many investigations including those in depression.

**Risk of Seizure:** The most serious risk associated with the use of rTMS is seizure. Since the adoption and widespread use of standard safety guidelines in 1997 [60], there has only been one documented seizure. The risk of seizure has been estimated to be less than 0.1% which is lower than the risk of seizure associated with pharmacologic antidepressants [61]. The risk of seizure is related to the various stimulation parameters (intensity, frequency, train duration), location of application, pre-existing risk of seizure, and substance/medication factors. In the very rare event a seizure is caused, removing the coil is typically sufficient to stop the seizure, and there is no increased risk of subsequent seizure. In order to mitigate the risk of seizure we will carefully individualize the intensity of stimulus (by performing a resting motor threshold determination), treat using standard treatment protocols (used safely in other studies) and minimize the use of any seizure threshold lowering medications. There is a significant comorbidity of epilepsy in the autism population, and as a population are at higher risk for seizure in general. However, in all rTMS studies performed to date on patients with Autism, there have been no seizures reported (Oberman 2016). The inclusion of patients with a known history of seizure in this study, also presents an increased risk above the general population. However, rTMS has been safely used in research and in general clinical use on patients with history of seizures. There have in fact been multiple studies explicitly using rTMS as a potential treatment modality for uncontrolled epilepsy (including refractory) - incidence of seizure in these studies remained between 1-2%, with only one seizure reported as atypical [62]. Overall, rTMS has been found to be safe for use in patients with known seizure disorders [63, 64]. Risk to participants with history of seizures will be minimized by exclusion of any participant who has experienced a seizure within the 6 months prior to enrollment. Participants will also remain on any anti-epileptic medication they were on prior to enrollment for the entire course of study.

**Risk of Site discomfort and headache:** Two relatively common risks associated with the use of rTMS include the risk of mild transient site discomfort during treatment (most patients), and the risk of headache (Approximately 5%) following treatment. Both of these potential side effects are typically mild. In terms of mitigating site discomfort, we will slowly ramp up stimulation intensity during the first three sessions. In our experience both clinically and experimentally this is a successful strategy. Additionally, due to the anti-pain effect of rTMS participants rapidly adjust to stimulation. In the unusual circumstance that a headache is caused by rTMS, over the counter analgesics are sufficient to alleviate the headaches, and will be recommended to all participants prior to start of treatment for use if they do experience a headache.

**Potential hearing loss:** The discharge of the TMS coil generates a high-energy click that may cause cochlear damage. Humans exposed to TMS have shown temporary increases in auditory threshold (especially at high frequencies) lasting at least 5 minutes and less than 4 hours. Foam earplugs can protect against these changes and will be worn during TMS sessions.

**Potential risks of fMRI paradigm:** The risks are minimal, but include the following: (a) Participants may experience boredom or sleepiness due to the monotony of the tasks and the number of repetitions of a task needed to obtain reliable results. (b) Eye strain and dry eyes may occur.

For each of these risks, precautions are taken to minimize these risks. (a) To minimize boredom or sleepiness experienced during the experiment participants are encouraged to arrive for the experiment well rested. For training and practice sessions, rest breaks are provided every 10 minutes as the procedure will allow (b) To minimize eye strain and dry eyes, participants will be told of this potential risk when scheduled for an appointment, so that they can use eye drops before the experiment, if necessary.

**Safety in the case of pregnancy:** This protocol will exclude pregnant women. Pregnancy status will be confirmed using a urine pregnancy test as part of the enrollment process.

**Confidentiality:** Every effort will be made to maintain participant confidentiality. Section 12 details how participant data will be managed securely, both in electronic and paper form. Participants who provide informed consent for the study have the option of allowing us to keep their information on file for up to 6 years, or they may opt to have their information destroyed after 1 year of having participated. No unauthorized personnel are allowed in the TMS or MRI areas while a subject is participating in a study.

## 16.0 Potential Benefits to Subjects or Others

This study has significant potential benefit to both directly to participants as well as indirect benefits to the entire autism population. Potential benefits for the participants are 1) reduction or remission of depression symptoms and 2) reduction of autism symptoms. While there are small risks associated with this study, they are mostly either mild (e.g. headache) or transient (seizure), and the risk of the latter is very low. In comparison with the potential benefit to improved quality of life and improved functioning for the participant, these risks are felt to be acceptable. Additionally, there is significant potential indirect benefit to all persons affected by autism by increasing our knowledge and understanding of the disorder and the further development of a potential treatment. This benefit is of particular importance due to general lack of treatment options which currently exist for the core symptoms of autism, and the known difficulty in treating depression in autism.

## 17.0 Sharing of Results with Subjects

Clinical scale results for any individual participant will be made available to that participant upon their request for their personal use after signing a release form. Also upon request, volunteers will receive a CD of their brain images which will be sent to them at a later date with release form. No results from one participant will be shared with a different participant. Any information critical to the safety of the participant (e.g. patient report of suicidal ideation or incidence of seizure) will be reported to appropriate emergency health care providers.

## 18.0 Drugs or Devices (if applicable)

**TMS device:** The TMS device that will be used for all rTMS procedures is Neuronetics/Neotonus Neopulse rTMS device with either a Neuronetics or Neopulse solid-coil head coil. Both coils are

butterfly coils, and the magnetic firing properties of both are identical when measured by an oscilloscope. The device is owned and maintained by the Brain Stimulation Department and is kept in a locked room when not in use. The device will only be operated by investigators and staff with both general clinical training and training specific to appropriate device operation. This device is available for use to multiple different research groups, and sessions will be scheduled for use using a department-wide scheduling system. Preference for scheduling time will be given to any other studies which are externally funded. Should the device require repair during the study, all effort will be made to seek access to an alternative but comparable TMS device, and appropriate approval will be sought for the change prior to any treatments on a different device. This is an FDA-approved device and is not considered investigational in its use in this study.

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